514 Rec'd PCT/PTO 28 FEB 2000

FORM-PTO-1390 (Rev. 10-96) U.S. DEPARTMENT OF COMMERCE PATENT AND TRADEMARK OFFICE

TRANSMITTAL LETTER TO THE UNITED STATES DESIGNATED/ELECTED OFFICE (DO/EO/US) CONCERNING A FILING UNDER 35 U.S.C. 371

ATTORNEY'S DOCKET NUMBER: 032929-001

U.S. APPLICATION NO. (If known, see 37 C.F.R. 1.5): 09/486394

INTERNATIONAL APPLICATION NO.	INTERNATIONAL FILING DATE	PRIORITY DATE CLAIMED
PCT/EP98/04773	30 July 1998	27 August 1997

TITLE OF INVENTION: DIAGNOSTIC KIT FOR SKIN TESTS, AND METHOD

APPLICANT(S) FOR DO/EO/US: Reinhard HÖPFL

Applicant herewith submits to the United States Designated/Elected Office (DO/EO/US) the following items and other information:

1. [X] This is a **FIRST** submission of items concerning a filing under 35 U.S.C. 371.
2. [ ] This is a **SECOND** or **SUBSEQUENT** submission of items concerning a filing under 35 U.S.C. 371.
3. [X] This is an express request to begin national examination procedures (35 U.S.C. 371(f)) at any time rather than delay examination until the expiration of the applicable time limit set in 35 U.S.C. 371(b) and the PCT Articles 22 and 39(1).
4. [X] A proper Demand for International Preliminary Examination was made by the 19th month from the earliest claimed priority date.
5. [X] A copy of the International Application as filed (35 U.S.C. 371(c)(2))
   a. [ ] is transmitted herewith (required only if not transmitted by the International Bureau).
   b. [X] has been transmitted by the International Bureau.
   c. [ ] is not required, as the application was filed in the United States Receiving Office (RO/US)
6. [X] A translation of the International Application into English (35 U.S.C. 371(c)(2)).
7. [X] Amendments to the claims of the International Application under PCT Article 19 (35 U.S.C. 371(c)(3))
   a. [ ] are transmitted herewith (required only if not transmitted by the International Bureau).
   b. [ ] have been transmitted by the International Bureau.
   c. [X] have not been made; however, the time limit for making such amendments has NOT expired.
   d. [ ] have not been made and will not be made.
8. [ ] A translation of the amendments to the claims under PCT Article 19 (35 U.S.C. 371(c)(3)).
9. [ ] An oath or declaration of the inventor(s) (35 U.S.C. 371(c)(4)).
10. [ ] A translation of the annexes to the International Preliminary Examination Report under PCT Article 36 (35 U.S.C. 371(c)(5)).

Items 11. to 16. below concern other document(s) or information included:

11. [X] An Information Disclosure Statement under 37 CFR 1.97 and 1.98.
12. [ ] An assignment document for recording. A separate cover sheet in compliance with 37 CFR 3.28 and 3.31 is included.
13. [X] A FIRST preliminary amendment.
    [ ] A SECOND or SUBSEQUENT preliminary amendment.
14. [ ] A substitute specification.
15. [ ] A change of power of attorney and/or address letter.
16. [ ] Other items or information:

514 Rec'd PCT/PTO 28 FEB 2000

U.S. APPLICATION NO. (If known, see 37 C.F.R. 1.5) 09/486394	INTERNATIONAL APPLICATION NO. PCT/EP98/04773	ATTORNEY'S DOCKET NUMBER 032929-001

17. ☒ The following fees are submitted:	CALCULATIONS	PTO USE ONLY
Basic National Fee (37 CFR 1.492(a)(1)-(5)):		
Search Report has been prepared by the EPO or JPO $840.00 (970)		
International preliminary examination fee paid to USPTO (37 CFR 1.482) $670.00 (956)		
No international preliminary examination fee paid to USPTO (37 CFR 1.482) but international search fee paid to USPTO (37 CFR 1.445(a)(2)) $690.00 (958)		
Neither international preliminary examination fee (37 CFR 1.482) nor international search fee (37 CFR 1.445(a)(2)) paid to USPTO $970.00 (960)		
International preliminary examination fee paid to USPTO (37 CFR 1.482) and all claims satisfied provisions of PCT Article 33(2)-(4) $96.00 (962)		
ENTER APPROPRIATE BASIC FEE AMOUNT =	$ 840.00	
Surcharge of **$130.00 (154)** for furnishing the oath or declaration later than 20 ☐ 30 ☐ months from the earliest claimed priority date (37 CFR 1.492(e)).	$	

Claims	Number Filed	Number Extra	Rate		
Total Claims	12 -20 =	0	X$18.00 (966)	$ --	
Independent Claims	1 -3 =	0	X$78.00 (964)	$ --	
Multiple dependent claim(s) (if applicable)			+$260.00 (968)	$	
TOTAL OF ABOVE CALCULATIONS =				$ 840.00	
Reduction for 1/2 for filing by small entity, if applicable. Verified Small Entity statement must also be filed. (Note 37 CFR 1.9, 1.27, 1.28).				$	-
SUBTOTAL =				$ 840.00	
Processing fee of **$130.00 (156)** for furnishing the English translation later than 20 ☐ 30 ☐ months from the earliest claimed priority date (37 CFR 1.492(f)). +				$	
TOTAL NATIONAL FEE =				$ 840.00	
Fee for recording the enclosed assignment (37 CFR 1.21(h)). The assignment must be accompanied by an appropriate cover sheet (37 CFR 3.28, 3.31). **$40.00 (581)** per property +				$	
TOTAL FEES ENCLOSED =				$ 840.00	
				Amount to be: refunded	$
				charged	$

a. ☒ A check in the amount of $ 840.00 to cover the above fees is enclosed.

b. ☐ Please charge my Deposit Account No. 02-4800 in the amount of $_____ to cover the above fees. A duplicate copy of this sheet is enclosed.

c. ☒ The Commissioner is hereby authorized to charge any additional fees which may be required, or credit any overpayment to Deposit Account No. 02-4800. A duplicate copy of this sheet is enclosed.

NOTE: Where an appropriate time limit under 37 CFR 1.494 or 1.495 has not been met, a petition to revive (37 CFR 1.137(a) or (b)) must be filed and granted to restore the application to pending status.

SEND ALL CORRESPONDENCE TO:

Peter K. Skiff
BURNS, DOANE, SWECKER & MATHIS, L.L.P.
P.O. Box 1404
Alexandria, Virginia 22313-1404

SIGNATURE

Teresa Stanek Rea
NAME

30,427
REGISTRATION NUMBER

Patent
Attorney's Docket No. 032929-001

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Patent Application of	)	
	)	
Reinhard HÖPFL et al	)	Group Art Unit: Unassigned
	)	
Application No.: Unassigned	)	
(Corresponds to PCT/EP98/04773)	)	
	)	
International Filing Date: 8 July 1998	)	Examiner: Unassigned
	)	
For: DIAGNOSTIC KIT FOR SKIN TESTS, AND METHOD	)	

PRELIMINARY AMENDMENT

BOX PCT
Assistant Commissioner for Patents
Washington, D.C. 20231

Sir:

Prior to examination, please amend the above-captioned application as follows:

IN THE CLAIMS:

Kindly amend the claims as follows:

Claim 3, line 1, delete "or 2".

Claim 4, line 1, delete "any of the preceding claims" and insert --claim 1--.

Claim 5, line 1, delete "any of the preceding claims" and insert --claim 1--.

Claim 7, line 1, delete "any of the preceding claims" and insert --claim 1--.

Claim 8, line 1, delete "any of the preceding claims" and insert --claim 1--.

Claim 11, line 4, delete "any of claims 1 to 9" and insert --claim 1--.

REMARKS

Entry of the foregoing amendments is respectfully requested.

The claims have been amended to eliminate multiple dependency and to place them in better condition for U.S. patent practice.

Should the Examiner have any questions concerning the subject application, a telephone call to the undersigned would be appreciated.

Respectfully submitted,

BURNS, DOANE, SWECKER & MATHIS, L.L.P.

By: _____
Teresa Stanek Rea
Registration No. 30,427

P.O. Box 1404
Alexandria, Virginia 22313-1404
(703) 836-6620

Date: February 28, 2000

M7124-PCT

Diagnosis kit for skin test and method for carrying out the same

Description

The present invention concerns a skin test for determining the immune status against human papilloma viruses.

Human papilloma viruses (HPV) are closely connected with malignant carcinomas of the anogenital tract, especially the neck of the uterus, the vulva, the penis and the anal duct. "High-risk" HPV types such as HPV 16 transform keratinocytes by means of the viral oncoproteins E6 and E7 contained therein by inactivating tumor suppressor proteins. It is assumed that the two oncoproteins E6 and E7 cooperatively contribute to the immortalizing (Hawley-Nelson, EMBO J., 8, 3905 (1989)).

The cancer cells transformed by HPV express the viral proteins E6 and E7 even in advanced tumor stages. It is assumed that a spontaneous or vaccination-induced immune reaction against these proteins could result in a regression of tumors caused by HPV. Until now, patients showing a spontaneous regression of their disease have not been examined due to the fact that examinations of the cellular defensive reaction *in vitro* are extremely time-consuming and, nevertheless, show a relatively low sensitivity. The role of the cellular immunological response in the case of vaccinations against HPV associated lesions is unclear.

Skin tests are partly already used in the clinical field in allergiology and for infectious diseases (mainly for clarifying tuberculosis in the form of the known TINE test).

There was, however, practically no specific application for virological questions or for cancer research until now.

Skin tests are also an excellent means for scientific questions and suitable for rapidly and practicably detecting complex cellular immune reactions *in vivo*. A skin test for examining the cellular immune reaction against "high risk" HPV is already known in prior art. This first application of the skin test for determining an immune reaction against a cancer associated virus was carried out with the coat protein L1 of the "high risk" HPV type 16 (utility model G9106105.9) and thereafter applied in animal models.

There is also a model for the skin test for the mouse; the immune reaction to a transplanted antigen was tested in this case (Mc Lean et al., J. Gen. Virol. **74**, 239-245 (1993)).

This known skin test did not detect the humoral immunological response to an HPV infection. In contrast to the cellular defense, the detection of the humoral immune reaction can be carried out relatively easiliy in the laboratory by means of serological tests. It is considered to be proven that antibodies against three-dimensionally intact structures of the virus coat proteins can neutralize papilloma viruses (Christensen et al. J Gen Virol **75**, 2271 (1994)). Antibodies against the early proteins E6 and E7 are associated with malignant tumors of the neck of the uterus (Jochmus-Kudielka et al. J. Natl. Cancer Inst. 81, 1698 (1989)).

A prophylactic vaccination with "virus like particles" consisting of coat proteins would preventively reduce an HPV infection and the risk of cancer associated therewith. The humoral immunological response against HPV, however, does not play a role with respect to the regression of already existing

lesions. The serology has therefore an epidemiological significance.

In summary, the experiments with animal models and female patients show that cellular immune reactions against viral proteins are detectable by skin tests. As a result of the data, it can be assumed that an immune reaction against the coat protein L1 is not associated with regression, as in advanced lesions intact virus particles cannot be formed any more. Thus, the transforming virus proteins E6 and E7 are the probable goal of a "healing" immune reaction. These tumor antigens are thus potential candidates for the development of a vaccine if, in addition to the prophylactic effectiveness, a therapeutic effectiveness is to be achieved by virus coat proteins. It is assumed that such a vaccination causes an activation of cytotoxic T cells and T helper cells against persistently infected genital lesions.

However, an *in vivo* test for detecting the success of such a vaccination against E6 and/or E7 by means of the cellular defensive reaction of the immunological system as a reaction to the vaccination has not been known until now.

Therefore, the object to be solved by the invention is to provide a test by means of which the immune status against E6 and/or E7 proteins of papilloma viruses can easily be detected *in vivo*. According to the invention, this object is solved by providing the diagnosis kit for skin tests according to independent claim 1 and the process for carrying out a skin test according to independent claim 10. Further advantageous embodiments and aspects of the invention are evident from the dependent claims and the description.

The skin test according to the invention can also be used for detecting spontaneous regressions of papilloma virus carcinoma precursors (CIN).

In a further aspect, the skin test according to the invention can also be used in the field of research in order to examine mechanisms in the regression of lesions caused by HPV.

In the following, the present invention is explained by means of an example, wherein the suitability of the skin test in principle for examining vaccination successes is shown with female patients who have not been vaccinated but partly showed spontaneous regressions of the cervix carcinomas and precursors thereof and thus a characteristic which is equivalent to the vaccination success. Synthetic peptides were used for the skin test against HPV16-E6 or -E7. Compared to the use of native proteins or fusion proteins, they have the advantage that a contamination with DNA could be excluded because it is absolutely certain that synthetic peptides are free from any problematic contamination with DNA.

An exemplary skin test with the peptide antigens for the oncoprotein HPV16-E7 was carried out on patients. Surprisingly, it could be shown that firstly, it is possible to detect a cellular immune reaction against the viral oncoprotein HPV16-E7 *in vivo*, and secondly, synthetic peptides can replace proteins for skin tests. Thirdly, an association of a positive skin test reaction in the sense of a "delayed type hypersensitivity" with regression of cervical cancer precursors could be observed. Such a correlation has not been previously known. The observation substantiates the potential significance of the antigen HPV16-E7 in a therapeutically effective vaccine and shows the applicability of the skin test according to the invention for determining the immune status with respect to E6 and E7.

For carrying out the example according to the invention, use was made of five relatively long peptides of 30 amino acids each, said peptides overlapping each other, for the oncoprotein HPV16-E7, and a control peptide of random sequence of the same length. The sequence for the 98 amino acids of the HPV16-E7 protein is known (Seedorf et al. J. Gen. Virol., **71**, 2719 (1990)). The peptides were produced at the Dep. IHB/AZL (Leiden, Holland) in a "multiple peptide synthesizer". The pepdtides were synthesized with the help of the "Fmoc" technology, precipitated by means of ether from trifluoro acid and subsequently lyophilized. The purity of the peptides was checked by means of "reverse phase high pressure liquidchromatography". Peptides for use with the skin test of the invention can, however, also be produced by any other way which is known to experts.

The sequences stated in a single letter code for the individual peptides tested were:

1. MHGDTPTLHEYMLDLQPETTDLYCYEQLND
2. DLYCYEQLNDSSEEEDEIDGPAGQAEPDRA
3. PAGQAEPDRAHYNIVTFCCKCDSTLRLCVQ
4. CDSTLRLCVQSTHVDIRTLEDLLMGTLGIV
5. STHVDIRTLEDLLMGTLGIVCPICSQKP

and as the control peptide:

6. SENKELKKAIDGLQGLLLGLRQRIETLEGK

The single letter code of the amino acids is, for example, explained in Römpps Chemie Lexikon, published by Georg Thieme Verlag, Stuttgart, volume 1, pages 160/161, (1989).

For the skin test, after a sterility test and a pyrogenic test, the peptides were solved in a concentration of 1 mg/ml in 70% glycerin and about 0.01 ml of this solution were injected in a strictly intracutaneous manner into the upper skin and the epidermis of the test person. Each Peptide was used separately (5 HPV16-E7 peptides + 1 control peptide). Of course, however, also combinations of the peptides among themselves and/or with respective peptides of HPV E6 can be used. Parallel to the skin test with E7 antigens, in most cases the general defensive condition against classical recall antigens was determined with an immune block, for example Mérieux Multitest "Sero", test system with 7 antigens and a control for determining the status of the cell-mediated immunity, produced by the Serotherapeutisches Institut Wien GmbH under license of Pasteur Merieux S.V., Lyon, France. Reactions with a reddish formation of papules with a diameter of at least 2 mm were evaluated as being positive.

The skin test was carried out on altogether 19 female patients with premalignant or malignant HPV caused lesions. The probability that such diseases are caused by the HPV type 16 is about 50%; in further 40% of the cases, other "high risk" HPV types have to be assumed:

Twelve female patients with relatively severe cervical intra-epithelial formations of neoplasms (ZIN III), 4 of them with a regression indication at the time of the examination (spontaneous regression of Pap III D to Pap II)
Seven female patients with cervix carcinomas.

Thus, 19 female patients were tested, 4 of which were regressors, and 15 of which were progressors.

Two men without any indication regarding HPV16 infection served as control persons.

In addition, in a manner commonly known to experts, the skin-tested persons were examined serologically by means of ELISA with "virus like particles (VLPs)" as an antigen with respect to neutralizing antibodies against HPV 16. Serologic examinations are not required for the specific skin test itself, but were carried out in the example as well to increase the scientific meaninfulness of the results. Smear or biopsy material of the lesions was stored for later virus typing by means of PCR at -80°C; first, examinations were carried out with dot blot (ViraType, Digene Diagnostics, Silver Spring, MD) with respect to HPV-6/11, -16/18 and -31/33/35. A lymphocyte bank was established for additional *in vitro* tests.

The female regressors (4/4) showed a slight (in 1 case) to strong (in 3 cases) immune reaction to individual peptides of the oncoprotein E7. The 15 female progressors and the two control persons did not show a clear reactivity in the skin test. Compared to a classical tuberculin reaction, the course of the skin test reaction was clearly delayed. Therefore, the best time for readings was in most cases one week after the testing. Reactions to the control peptide were not found in any case.

The reactions were photodocumented, one reaction was biopsied and examined histologically. Similar to the observations already made in former skin tests with L1, a lymphocytic infiltrate was found, which seemed to be compatible with a "delayed type hypersensitivity reaction". Antibodies against HPV16-L1 were found in 4 of the 7 female patients with a cervix carcinoma and only in one female patient with CIN. In summary, thus the antibody detection against HPV16-L1-VLPs was obviously more associated with a progression, whereas a cellular immune reaction against HPV16-E7 in the skin test was

associated with the regression of precancerous cervical lesions.

The results prove the effectiveness of the skin test for a sensitive detection of a cellular immune reaction against HPV16-E7, which, in the cases examined, was associated with a spontaneous regression of an HPV associated cancer precursor lesion.

As, by means of the skin test described, an immune reaction was detected which was associated with regression of cancer precursor lesions, the skin test can be used as an instrument for controlling a vaccination success against E6 and/or E7. A broad screening by means of skin testing for the clarification of the mechanisms of an immunologically mediated tumor regression is also possible. Because of the extremely high expenditure for *in vitro* experiments, it is hardly possible to test a cellular immunity for a large number of patients by means of laboratory tests. In future studies regarding vaccination with a hypothetically therapeutically effective vaccine against the cervix carcinoma by immunisation against the tumor protein HPV16-E7, the skin test according to the invention is, because it is practicable and sensitive, an ideal method for detecting the desired reaction of the cellular immunological system to the vaccination.

In the example mentioned above, the peptides used were dissolved in 70% glycerin. Glycerin is preferred because it has a viscosity suitable for an intracutaneous application and, in addition, has a disinfecting effect. According to the invention, however, other suitable solvents can also be used; possibly, due to the solubility characteristics of a peptide, it has to be dissolved in a specially adapted solution. Further additives such as emulsifiers, chelating agents,

disinfectants and others can also be added to the solvents used according to the invention.

The skin test can be effected by an intracutaneous injection of an effective amount of dissolved E6 and/or E7 antigen by means of a syringe. Accordingly, the diagnosis kit would contain on or more ampoules, syringes and cannulas. According to the invention, the use of applicators specially adapted for skin tests is also possible and preferred, for example the multitest "SERO" test stamp of Sero-Mérieux (see above) or the stamp for the TINE test. Accordingly, the diagnosis kit according to the invention can contain ampoules with antigen(s) and/or applicators.

In the example described above, the way the skin test of the invention works was described with HPV16-E7. However, E6 can be used as well. It is also possible to use the skin test by using E6 and/or E7 of other HPV types for detecting an immunological response with respect to these strains. Because of the close degree of relationship of different HPV types, it is finally also possible to make use of cross-reactions between different HPV types to introduce a more universal skin test against different HPV types at the same time.

The amount of antigens which has to be applied for a visible immune reaction depends on various factors. Accordingly, according to the invention, it can range between 0.01 and 10 µg, preferably between 0.05 and 5 µg antigen per application. In this case, when preparing antigen solutions for diagnosis kits, it has to be taken into account that only part of the solution will get into the intracutaneous region, whereas another part can be lost on the skin of the test person or will remain in the solution ampoule.

Diagnosis kit for skin test and method for carrying out the same

Claims

1. A skin test diagnosis kit for detecting an immune reaction against the oncoprotein E6 and/or E7 of a human papilloma virus type, said diagnosis kit containing an effective amount of the oncoprotein E6 and/or E7 and/or at least an immunologically effective portion of E6 and/or E7 of a human papilloma virus type.

2. The diagnosis kit of claim 1, characterized in that the contained portion of the oncoprotein is derived from HPV16.

3. The diagnosis kit of claim 1 or 2, characterized in that the immunologically effective portion of the human papilloma virus type is at least one synthetically produced peptide.

4. The diagnosis kit of any of the preceding claims, characterized in that the contained oncoprotein E7 or the immunologically effective portion thereof is or are from HPV16.

5. The diagnosis kit of any of the preceding claims, characterized in that the contained oncoprotein or the immunologically effective portion thereof is dissolved in a solvent.

6. The diagnosis kit of claim 5, characterized in that the solvent is 70% glycerin.

7. The diagnosis kit of any of the preceding claims, characterized in that the amount of oncoprotein or the immunologically effective portion is 0.01 to 10 µg per charge to be applied.

8. The diagnosis kit of any of the preceding claims, characterized in that said diagnosis kit further comprises an applicator, by means of which said effective amount of the oncoprotein or the immunologically effective portion thereof can be injected intracutaneously.

9. The diagnosis kit of claim 8, characterized in that said applicator is a syringe.

10. The diagnosis kit of claim 8, characterized in that said applicator is a test stamp as used, for example, for the TINE test or the multitest "Sero".

11. A process for carrying out a skin test for detecting an immunological response with respect to the oncoproteins E6 and/or E7 of an HPV type, comprising the following steps:

a) providing a diagnosis kit of any of claims 1 to 9;

b) intracutaneous application of an effective amount of at least one oncoprotein E6 and E7 or effective portions thereof into a test person;

c) after a sufficient incubation time, visual inspection of the skin regions of the application to detect an immunological response.

12. The process of claim 11, characterized in that the visual inspection of the skin region takes place one week after application of the oncoprotein.

Diagnosis kit for skin test and method for carrying out the same

Abstract

A skin test for detecting the immune status with respect to the transforming virus proteins E6 and E7 of human papilloma viruses is provided. The skin test comprises a diagnosis kit containing an effective amount of the protein E6 and/or E7 or at least an immunologically effective portion of E6 and/or E7. The skin test is carried out by intracutaneously applying the protein E6/E7 of the diagnosis kit and visually inspecting the respective skin region to detect reddening.

SEQUENCE LISTING

(1) GENERAL INFORMATION

(i) APPLICANT:

(A) NAME: MediGene AG

(B) STREET: Lochhamer Str. 11

(C) CITY: Munich

(D) STATE: Bavaria

(E) COUNTRY: Germany

(F) ZIP: 81927

(G) TELEPHONE: +49-89-8956320

(H) FAX: +49-89-89563220

(A) NAME: Dr. Reinhard Hoepfl

(B) STREET: Anichstr. 35

(C) CITY: Innsbruck

(E) COUNTRY: Austria

(F) ZIP: 6020

(ii) TITLE OF INVENTION: Diagnosis kit for skin test and method for carrying out the same

(iii) NUMBER OF SEQUENCES: 6

(iv) COMPUTER READABLE FORM:

(A) MEDIUM TYPE: floppy disk

(B) COMPUTER: IBM PC compatible

(C) OPERATING SYSTEM: PC-DOS/MS-DOS

(D) SOFTWARE: PatentIn Release #1.0, version #1.30 (EPO)

(2) INFORMATION FOR SEQ ID NO: 1:

(i) SEQUENCE CHARACTERISTICS:

(A) LENGTH: 30 amino acids

(B) TYPE: amino acid

(C) STRANDENESS:

(D) TOPOLOGY: unknown

(ii) MOLECULE TYPE: peptide

(iii) HYPOTHETICAL: NO

(iv) ANTISENSE: NO

(v) FRAGMENT TYPE: inner fragment

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 1:

Met His Gly Asp Thr Pro Thr Leu His Glu Tyr Met Leu Asp Leu Gln
1 5 10 15

Pro Glu Thr Thr Asp Leu Tyr Cys Tyr Glu Gln Leu Asn Asp
20 25 30

(2) INFORMATION FOR SEQ ID NO: 2:

(i) SEQUENCE CHARACTERISTICS:

(A) LENGTH: 30 amino acids

(B) TYPE: amino acid

(C) STRANDENESS:

(D) TOPOLOGY: unknown

(ii) MOLECULE TYPE: peptide

(iii) HYPOTHETICAL: NO

(iv) ANTISENSE: NO

(v) FRAGMENT TYPE: inner fragment

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 2:

Asp Leu Tyr Cys Tyr Glu Gln Leu Asn Asp Ser Ser Glu Glu Glu Asp
1 5 10 15

Glu Ile Asp Gly Pro Ala Gly Gln Ala Glu Pro Asp Arg Ala
20 25 30

(2) INFORMATION FOR SEQ ID NO: 3:

(i) SEQUENCE CHARACTERISTICS:

(A) LENGTH: 30 amino acids

(B) TYPE: amino acid

(C) STRANDENESS:

(D) TOPOLOGY: unknown

(ii) MOLECULE TYPE: peptide

(iii) HYPOTHETICAL: NO

(iv) ANTISENSE: NO

(v) FRAGMENT TYPE: inner fragment

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 3:

Pro Ala Gly Gln Ala Glu Pro Asp Arg Ala His Tyr Asn Ile Val Thr
1 5 10 15

Phe Cys Cys Lys Cys Asp Ser Thr Leu Arg Leu Cys Val Gln
20 25 30

(2) INFORMATION FOR SEQ ID NO: 4:

(i) SEQUENCE CHARACTERISTICS:

(A) LENGTH: 30 amino acids

(B) TYPE: amino acid

(C) STRANDENESS:

(D) TOPOLOGY: unknown

(ii) MOLECULE TYPE: peptide

(iii) HYPOTHETICAL: NO

(iv) ANTISENSE: NO

(v) FRAGMENT TYPE: inner fragment

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 4:

Cys Asp Ser Thr Leu Arg Leu Cys Val Gln Ser Thr His Val Asp Ile
1 5 10 15

Arg Thr Leu Glu Asp Leu Leu Met Gly Thr Leu Gly Ile Val
20 25 30

(2) INFORMATION FOR SEQ ID NO: 5:

(i) SEQUENCE CHARACTERISTICS:

(A) LENGTH: 28 amino acids

(B) TYPE: amino acid
(C) STRANDENESS:
(D) TOPOLOGY: unknown
(ii) MOLECULE TYPE: peptide
(iii) HYPOTHETICAL: NO
(iv) ANTISENSE: NO
(v) FRAGMENT TYPE: inner fragment
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 5:

Ser Thr His Val Asp Ile Arg Thr Leu Glu Asp Leu Leu Met Gly Thr
1 5 10 15

Leu Gly Ile Val Cys Pro Ile Cys Ser Gln Lys Pro
20 25

(2) INFORMATION FOR SEQ ID NO: 6:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 30 amino acids
(B) TYPE: amino acid
(C) STRANDENESS:
(D) TOPOLOGY: unknown
(ii) MOLECULE TYPE: peptide
(iii) HYPOTHETICAL: NO
(iv) ANTISENSE: NO
(v) FRAGMENT TYPE: inner fragment
(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 6:

Ser Glu Asn Lys Glu Leu Lys Lys Ala Ile Asp Gly Leu Gln Gly Leu
1 5 10 15

Leu Leu Gly Leu Arg Gln Arg Ile Glu Thr Leu Glu Gly Lys
20 25 30

COMBINED DECLARATION FOR PATENT APPLICATION AND POWER OF ATTORNEY (Includes Reference to Provisional and PCT International Applications)	Attorney's Docket No. 032929-001

As a below named inventor, I hereby declare that:
My residence, post office address and citizenship are as stated below next to my name;
I believe I am the original, first and sole inventor (if only one name is listed below) or an original, first and joint inventor (if plural names are listed below) of the subject matter which is claimed and for which a patent is sought on the invention entitled:

DIAGNOSTIC KIT FOR SKIN TESTS, AND METHOD

the specification of which (check only one item below):

☐ is attached hereto.

☐ was filed as United States application
Number _____
on _____
and was amended
on _____ (if applicable).

☒ was filed as PCT international application
Number PCT/EP98/04773
on 30 July 1998
and was amended
on _____ (if applicable).

I hereby state that I have reviewed and understand the contents of the above-identified specification, including the claims, as amended by any amendment referred to above.

I acknowledge the duty to disclose to the Office all information known to me to be material to patentability as defined in Title 37, Code of Federal Regulations, §1.56.

I hereby claim foreign priority benefits under Title 35, United States Code, §119 (a)-(e) of any foreign application(s) for patent or inventor's certificate or of any PCT international application(s) designating at least one country other than the United States of America listed below and have also identified below any foreign application(s) for patent or inventor's certificate or any PCT international application(s) designating at least one country other than the United States of America filed by me on the same subject matter having a filing date before that of the application(s) of which priority is claimed:

PRIOR FOREIGN/PCT APPLICATION(S) AND ANY PRIORITY CLAIMS UNDER 35 U.S.C. §119:

COUNTRY (if PCT, indicate "PCT")	APPLICATION NUMBER	DATE OF FILING (day, month, year)	PRIORITY CLAIMED UNDER 35 U.S.C. §119
DE	197 37 409.3	27 August 1997	X Yes _No
			_Yes _No
			_Yes _No
			_Yes _No
			_Yes _No

I hereby claim the benefit under Title 35, United States Code § 119(e) of any United States provisional application(s) listed below.

_____ _____
(Application Number) (Filing Date)

_____ _____
(Application Number) (Filing Date)

COMBINED DECLARATION FOR PATENT APPLICATION AND POWER OF ATTORNEY (CONT'D) (Includes Reference to Provisional and PCT International Applications)	Attorney's Docket No. 032929-001

I hereby claim the benefit under Title 35, United States Code, §120 of any United States applications(s) or PCT international application(s) designating the United States of America that is/are listed below and, insofar as the subject matter of each of the claims of this application is not disclosed in that/those prior application(s) in the manner provided by the first paragraph of Title 35, United States Code, §112, I acknowledge the duty to disclose to the Office all information known to me to be material to the patentability as defined in Title 37, Code of Federal Regulations §1.56, which became available between the filing date of the prior application(s) and the national or PCT international filing date of this application:

PRIOR U.S. APPLICATIONS OR PCT INTERNATIONAL APPLICATIONS DESIGNATING THE U.S. FOR BENEFIT UNDER 35 U.S.C. §120:

U.S. APPLICATIONS		STATUS *(check one)*		
U.S. APPLICATION NUMBER	U.S. FILING DATE	PATENTED	PENDING	ABANDONED

PCT APPLICATIONS DESIGNATING THE U.S.					
PCT APPLICATION NO.	PCT FILING DATE	U.S. APPLICATION NUMBERS ASSIGNED (if any)			

I hereby appoint the following attorneys and agent(s) to prosecute said application and to transact all business in the Patent and Trademark Office connected therewith and to file, prosecute and to transact all business in connection with international applications directed to said invention:

William L. Mathis	17,337	R. Danny Huntington	27,903	Gerald F. Swiss	30,113
Robert S. Swecker	19,885	Eric H. Weisblatt	30,505	Michael J. Ure	33,089
Platon N. Mandros	22,124	James W. Peterson	26,057	Charles F. Wieland III	33,096
Benton S. Duffett, Jr.	22,030	Teresa Stanek Rea	30,427	Bruce T. Wieder	33,815
Norman H. Stepno	22,716	Robert E. Krebs	25,885	Todd R. Walters	34,040
Ronald L. Grudziecki	24,970	William C. Rowland	30,888	Ronni S. Jillions	31,979
Frederick G. Michaud, Jr.	26,003	T. Gene Dillahunty	25,423	Harold R. Brown III	36,341
Alan E. Kopecki	25,813	Patrick C. Keane	32,858	Allen R. Baum	36,086
Regis E. Slutter	26,999	Bruce J. Boggs, Jr.	32,344	Steven M. du Bois	35,023
Samuel C. Miller, III	27,360	William H. Benz	25,952	Brian P. O'Shaughnessy	32,747
Robert G. Mukai	28,531	Peter K. Skiff	31,917	Kenneth B. Leffler	36,075
George A. Hovanec, Jr.	28,223	Richard J. McGrath	29,195	Fred W. Hathaway	32,236
James A. LaBarre	28,632	Matthew L. Schneider	32,814		
E. Joseph Gess	28,510	Michael G. Savage	32,596		

21839

and: _____

Address all correspondence to:

21839

Peter K. Skiff
BURNS, DOANE, SWECKER & MATHIS, L.L.P.
P.O. Box 1404
Alexandria, Virginia 22313-1404

Address all telephone calls to: Peter K. Skiff at (703) 836-6620.

I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

COMBINED DECLARATION FOR PATENT APPLICATION AND POWER OF ATTORNEY (CONT'D) (Includes Reference to Provisional and PCT International Applications)	Attorney's Docket No. 032929-001

FULL NAME OF SOLE OR FIRST INVENTOR Reinhard HÖPFL	SIGNATURE	DATE 5.6.2000
RESIDENCE Universitatsklinik fur Dermatologie und Venerologie, Anichstrasse 35, A-6020 Innsbruch, AT	CITIZENSHIP AT	
POST OFFICE ADDRESS Universitatsklinik fur Dermatologie und Venerologie, Anichstrasse 35, A-6020 Innsbruch, AT		
FULL NAME OF SECOND JOINT INVENTOR, IF ANY	SIGNATURE	DATE
RESIDENCE	CITIZENSHIP	
POST OFFICE ADDRESS		
FULL NAME OF THIRD JOINT INVENTOR, IF ANY	SIGNATURE	DATE
RESIDENCE	CITIZENSHIP	
POST OFFICE ADDRESS		
FULL NAME OF FOURTH JOINT INVENTOR, IF ANY	SIGNATURE	DATE
RESIDENCE	CITIZENSHIP	
POST OFFICE ADDRESS		
FULL NAME OF FIFTH JOINT INVENTOR, IF ANY	SIGNATURE	DATE
RESIDENCE	CITIZENSHIP	
POST OFFICE ADDRESS		
FULL NAME OF SIXTH JOINT INVENTOR, IF ANY	SIGNATURE	DATE
RESIDENCE	CITIZENSHIP	
POST OFFICE ADDRESS		
FULL NAME OF SEVENTH JOINT INVENTOR, IF ANY	SIGNATURE	DATE
RESIDENCE	CITIZENSHIP	
POST OFFICE ADDRESS		
FULL NAME OF EIGHTH JOINT INVENTOR, IF ANY	SIGNATURE	DATE
RESIDENCE	CITIZENSHIP	
POST OFFICE ADDRESS		